

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody

Catalog # ABO13126

Specification

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format **Description** Anti-Phospho-HSF1 (S3 WB, IHC, IF, ICC, IP, FC <u>000613</u> Rabbit Rabbit IgG Human Monoclonal Liquid

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody . Tested in WB, IHC, ICC/IF, IP, Flow Cytometry applications. This antibody reacts with Human.

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody - Additional Information

Gene ID 3297

Other Names Heat shock factor protein 1, HSF 1, Heat shock transcription factor 1 {ECO:0000312|HGNC:HGNC:5224}, HSTF 1, HSF1 (HGNC:5224), HSTF1

Calculated MW 57260 MW KDa

Application Details WB 1:500-1:2000
IHC 1:50-1:200
ICC/IF 1:50-1:200
IP 1:50
FC 1:50

Subcellular Localization

Cytoplasm. Nucleus. Cytoplasmic during normal growth. On activation, translocates to nuclear stress granules. Colocalizes with SUMO1 in nuclear stress granules.

Contents Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.

Immunogen A synthesized peptide derived from human Phospho-HSF1 (S326)

Purification Affinity-chromatography

Storage

Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated



freeze-thaw cycles.

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody - Protein Information

Name HSF1 (<u>HGNC:5224</u>)

Synonyms HSTF1

Function

Functions as a stress-inducible and DNA-binding transcription factor that plays a central role in the transcriptional activation of the heat shock response (HSR), leading to the expression of a large class of molecular chaperones, heat shock proteins (HSPs), that protect cells from cellular insult damage (PubMed:11447121, PubMed:12659875, PubMed:12917326, PubMed:15016915, PubMed:18451878, PubMed:1871105, PubMed:1986252, PubMed:25963659, PubMed:26754925, PubMed:7623826, PubMed:7760831, PubMed:8940068, PubMed:8946918, PubMed:9121459, PubMed:9341107, PubMed:9499401, PubMed:9535852, PubMed:9727490). In unstressed cells, is present in a HSP90-containing multichaperone complex that maintains it in a non-DNA-binding inactivated monomeric form (PubMed:11583998, PubMed:16278218, PubMed:9727490). Upon exposure to heat and other stress stimuli, undergoes homotrimerization and activates HSP gene transcription through binding to site-specific heat shock elements (HSEs) present in the promoter regions of HSP genes (PubMed:10359787, PubMed:11583998. PubMed:12659875, PubMed:16278218, PubMed:1871105, PubMed:1986252, PubMed:25963659, PubMed:26754925, PubMed:7623826, PubMed:7935471, PubMed:8455624, PubMed:8940068, PubMed:9499401, PubMed:9727490). Upon heat shock stress, forms a chromatin-associated complex with TTC5/STRAP and p300/EP300 to stimulate HSR transcription, therefore increasing cell survival (PubMed:18451878). Activation is reversible, and during the attenuation and recovery phase period of the HSR, returns to its unactivated form (PubMed:<a href="http://www.uniprot.org/citations/11583998"



target="_blank">11583998, PubMed:16278218). Binds to inverted 5'-NGAAN-3' pentamer DNA sequences (PubMed:1986252, PubMed:26727489). Binds to chromatin at heat shock gene promoters (PubMed:25963659). Activates transcription of transcription factor FOXR1 which in turn activates transcription of the heat shock chaperones HSPA1A and HSPA6 and the antioxidant NADPH-dependent reductase DHRS2 (PubMed:34723967). Activates transcription of the heat shock chaperones HSPA1A and HSPA6 and the antioxidant NADPH-dependent reductase DHRS2 (PubMed:34723967). Also serves several other functions independently of its transcriptional activity. Involved in the repression of Ras-induced transcriptional activation of the c-fos gene in heat-stressed cells (PubMed:9341107). Positively regulates pre-mRNA 3'-end processing and polyadenylation of HSP70 mRNA upon heat-stressed cells in a symplekin (SYMPK)-dependent manner (PubMed:14707147). Plays a role in nuclear export of stress- induced HSP70 mRNA (PubMed:17897941). Plays a role in the regulation of mitotic progression (PubMed:18794143). Also plays a role as a negative regulator of non-homologous end joining (NHEJ) repair activity in a DNA damage-dependent manner (PubMed:26359349). Involved in stress-induced cancer cell proliferation in a IER5-dependent manner (PubMed:26754925).

Cellular Location

Nucleus. Cytoplasm. Nucleus, nucleoplasm. Cytoplasm, perinuclear region. Cytoplasm, cytoskeleton, spindle pole. Cytoplasm, cytoskeleton, microtubule organizing center, centrosome Chromosome, centromere, kinetochore Note=The monomeric form is cytoplasmic in unstressed cells (PubMed:26159920, PubMed:8455624). Predominantly nuclear protein in both unstressed and heat shocked cells (PubMed:10359787, PubMed:10413683). Translocates in the nucleus upon heat shock (PubMed:8455624). Nucleocytoplasmic shuttling protein (PubMed:26159920). Colocalizes with IER5 in the nucleus (PubMed:27354066). Colocalizes with BAG3 to the nucleus upon heat stress (PubMed:26159920, PubMed:8455624). Localizes in subnuclear granules called nuclear stress bodies (nSBs) upon heat shock (PubMed:10359787, PubMed:10747973, PubMed:11447121, PubMed:11514557, PubMed:19229036, PubMed:24581496, PubMed:25963659). Colocalizes with SYMPK and SUMO1 in nSBs upon heat shock (PubMed:10359787, PubMed:11447121, PubMed:11514557, PubMed:12665592, PubMed:14707147) Colocalizes with PRKACA/PKA in the nucleus and nSBs upon heat shock (PubMed:21085490). Relocalizes from the nucleus to the cytoplasm during the attenuation and recovery phase period of the heat shock response (PubMed:26159920). Translocates in the cytoplasm in a YWHAE- and XPO1/CRM1-dependent manner (PubMed:12917326). Together with histone H2AX, redistributed in discrete nuclear DNA damage-induced foci after ionizing radiation (IR) (PubMed:26359349). Colocalizes with calciumresponsive transactivator SS18L1 at kinetochore region on the mitotic chromosomes (PubMed:18794143). Colocalizes with gamma tubulin at centrosome (PubMed:18794143). Localizes at spindle pole in metaphase (PubMed:18794143). Colocalizes with PLK1 at spindle poles during prometaphase (PubMed:18794143).

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot



- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

Anti-Phospho-HSF1 (S326) Rabbit Monoclonal Antibody - Images



Figure 1. Western blot analysis of HSF1 using anti-HSF1 antibody (P00250).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,

Lane 2: human K562 whole cell lysates,

Lane 3: human HepG2 whole cell lysates,

Lane 4: human MCF-7 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-HSF1 antigen affinity purified monoclonal antibody (Catalog # P00250) at 1:500 overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:1000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for HSF1 at approximately 80 kDa. The expected band size for HSF1 is at 57 kDa.





Figure 2. IHC analysis of HSF1 using anti-HSF1 antibody (P00250).

HSF1 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-HSF1 Antibody (P00250) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



Figure 3. IHC analysis of HSF1 using anti-HSF1 antibody (P00250).

HSF1 was detected in a paraffin-embedded section of human spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-HSF1 Antibody (P00250) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.





Immunofluorescent analysis using the Antibody at 1:50 dilution.



Immunofluorescent analysis using the Antibody at 1:150 dilution.